

employment of the cDNA library of the above-stated tissues (e.g., brain, testis, heart, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. In detail, the solution 50 μ l containing 5 pmol of the each primer, 5 μ l of 100 mM Tris-HCl buffer (pH9.0), 5 μ l of 500 mM potassium chloride solution, 3 μ l of 25 mM magnesium chloride solution, 4 μ l of 2.5 mM deoxyribonucleotide solution, 1 μ l of cDNA solution, and 0.5 μ l of TaKaRa Tag™ was prepared. The PCR reaction was performed according to the following program, namely, the resultant solution was placed at 95°C for 1 minute in TaKaRa PCR Thermal Cycler MP (Takara shuzo Co., Ltd.), then at 95°C for 30 seconds, at 65°C for 1 minute and at 72°C for 2 minutes; this cycle was repeated 35 times in total and further reacted at 72°C for 10 minutes to obtain the target PCR fragment.

The paragraph beginning on page 59, line 15 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO: 17, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGCACAGATCAGAGCCATTCTGA (SEQ ID NO:33); and TTACAGTAGTGGCAGTAACACTTGG) (SEQ ID NO:34); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., epididymis, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 60, line 1 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:18, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGAAGTTCGTCCCCTGCCTCCTGC (SEQ ID NO:35); and

TCACCCTCGGAAGAAGCTGATGAGA) (SEQ ID NO:36); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., spinal cord, T cells, retina, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 60, line 21 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:19, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGGGACCTGTGCGGTTGGGAATAT (SEQ ID NO:37); and TCAAAGATCTTCTCGGTCAAGTTTG) (SEQ ID NO:38); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., cerebellum, adrenal, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 61, line 6 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:20, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGTTTGGCCCACTGAACTCATCC (SEQ ID NO:39); and TCATGAAAATATCCATTCTACCTTG) (SEQ ID NO:40); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., dendritic cells, T cells, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as

a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 61, line 26 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:21, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGGAAGCTGCTTCAAGTGACCATTC (SEQ ID NO:41); and TCAGTTCTTGGTTTTTCCTTGTGCA) (SEQ ID NO:42); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., vascular endothelial cells, bone marrow, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 61, line 11 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:22, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGCGACCCCAGGGCCCCGCCGCCT (SEQ ID NO:43); and TTATTTTGGTAGTTCTTCAATAATG) (SEQ ID NO:44); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., thymus, placenta, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 62, line 31 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:23, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGAAGTTACAGTGTGTTCCCTTT (SEQ ID NO:45); and TCAGGAGGCCGATGGGGGCCAGCAC) (SEQ ID NO:46); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., monocytes, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 63, line 16 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other- words, using the base sequence as described in SEQ ID NO:24, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGGCCAGCCTGGGGCTGCTGCTCC (SEQ ID NO:47); and TCATGAGGCTCCTGCAGAGGTCTGA) (SEQ ID NO:48); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., cerebellum, lung, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 64, line 1 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:25, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGAAACTCCTGCTGCTGGCTCTTC (SEQ ID NO:49); and TCATGAGCTATGGTGAACATTTGGA) (SEQ ID NO:50); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., epididymis, etc.)

or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 64, line 21 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:26, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGGGCGGCCTGCTGCTGGCTGCTT (SEQ ID NO:51); and CTAAGTGACAGGAAGCCCAGGCTC) (SEQ ID NO:52); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., epididymis, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 65, line 7 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:27, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGGCCCGGCATGGGTTACCGCTGC (SEQ ID NO:53); and TTACAGCTCCCCTGGCGGCCCGCCT) (SEQ ID NO:54); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., T cells, placenta, liver, large intestine, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 65, line 27 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:28, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGTGCTGGCTGCGGGCATGGGGCC (SEQ ID NO:55); and TTATCTATTCATCATATATTCTTA) (SEQ ID NO:56); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., testis, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 66, line 11 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:29, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGGGGTTCCCGGCCGCGGCGCTGC (SEQ ID NO:57); and CTACGCCGAGACCGTGGGCCTGCGG) (SEQ ID NO:58); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., pancreas, placenta, etc.) or RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 66, line 31 has been amended to read as follows:

The cDNA fragment could be obtained by conventional methods, based on the above information. In other words, using the base sequence as described in SEQ ID NO:30, the primers for PCR corresponding to 5' end and 3' end (e.g., ATGCGAGGTGGCAAATGCAACATGC (SEQ ID NO:59); and TCATAAACTTGTGTTGGGCTTTAGG) (SEQ ID NO:60); were prepared, followed by employment of the cDNA library of the above-stated tissues (e.g., placenta, etc.) or

RT-PCR with use of mRNA derived from the aforementioned tissues as a template to obtain the cDNA fragment. The reaction conditions to be applied to PCR were those as described in EXAMPLE 1.

The paragraph beginning on page 67, line 11 has been amended to read as follows:

First of all, the PCR was performed by using the synthetic DNA [5'-ACGCTCGAGTTACTTGTCATCGTCGTCCTTGTAGTCCGTATGGGGCATCTGCCCTTTTTC-3' : (SEQ ID NO:62)], which was designed to locate the synthetic DNA [5'-TCGGAATTCGCCATGGCCAAGTACCTGGCCCAGATC -3' : (SEQ ID NO:61)] having the recognition site for the restriction enzyme Eco RI immediately before the translation start codon initiator, the FLAG sequence consisting of 8 amino acids (Asp-Tyr-Lys-Asp-Asp-Asp-Asp-Lys) (SEQ ID NO:71); at C-terminus of the TGC-480 protein, and subsequent stop codon and the recognition site for the restriction enzyme Xho I, by using the cDNA fragment encoding the TGC-480 protein obtained in EXAMPLE 1 as a template. The PCR reaction was performed according the following program, with use of Pyrobest DNA Polymerase (Takara Shuzo Co., Ltd.); namely, the reaction mixture was placed at 94°C for 1 minute, then at 98°C for 10 seconds, at 60°C for 30 seconds and at 72°C for 1 minute; this cycle was repeated 25 times in total. Finally, extension reaction at 72°C for 10 minutes was performed to obtain the DNA fragment containing the ORF of TGC-480. The resultant DNA fragment was cleaved with the restriction enzyme Eco RI and Xho I, followed by insertion of them into Eco RI/Xho I sites in pCAN618 to obtain the expression vector for TGC-480 protein, pCAN618/TGC-480FLAG for animal cells.

The paragraph beginning on page 68, line 13, has been amended to read as follows:

The expression vector to express TGC-623 product in animal cells was obtained by insertion of the DNA fragment containing ORF encoding the TGC-623 product into the expression vector pCAN618FLAG for animal cells. pCAN618FLAG

was derived from the plasmid vector pCAN618, and pCAN618FLAG can express the target protein as the FLAG fused protein, by coinciding the reading frame of the base sequence encoding the FLAG sequence for 8 amino acids existing immediately after Sal I site (Asp-Tyr-Lys-Asp-Asp-Asp-Asp-Lys) (SEQ ID NO:71); to the end codon.

The paragraph beginning on page 69, line 10 has been amended to read as follows:

First of all, the PCR was performed by using the synthetic DNA [5'-ACGCTCGAGTTACTTGTCATCGTCGTCCTTG TAGTCTGAGGCTCCTGCAGAGGTCTGAGA-3': (SEQ ID NO:64)], which was designed to locate the FLAG sequence (Asp-Tyr-Lys-Asp-Asp-Asp-Asp-Lys) (SEQ ID NO:71); and subsequently the end codon as well as the recognition site for the restriction enzyme Xho I in this sequence order, and the synthetic DNA [5'-TCGGAATTCGCCATGGCCAGCCTGGGGCTGCTGCTC -3': (SEQ ID NO:63)] having as a template the recognition site for the restriction enzyme Eco RI immediately before the translation start codon initiator, by using the cDNA fragment encoding TGC-711 protein obtained in EXAMPLE 9 as a template. The PCR reaction and subsequent treatments were performed under the similar conditions as those in EXAMPLE 16, to obtain the expression vector for human TGC-711 protein, or pCAN618/TGC-711 FLAG for animal cells. This expression vector was introduced into COS7 cells in the similar manners for those in EXAMPLE 16, and the culture supernatant was prepared, thereby being used for performing the Western Blot analysis. As the result, it was revealed that TGC-711 protein was secreted into the culture supernatant (Fig. 1).

The paragraph beginning on page 69, line 10 has been amended to read as follows:

First of all, the PCR was performed by using the synthetic DNA [5'-ACGCTCGAGTTACTTGTCATCGTCGTCCTTG TAGTCTGAGCTATGGTGAA